Seroprevalence of bluetongue serotype 8 in cattle in the Netherlands in spring 2007, and its consequences

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A cross-sectional study was carried out in spring 2007, at the end of the first bluetongue outbreak season, to determine the geographical spread of bluetongue virus serotype 8 (BTV-8) infection in cattle in the Netherlands and the consequences for some production parameters. Blood samples from cattle submitted to the laboratory of the Dutch Animal Health Service for other voluntary and obligatory health programmes were tested serologically for BTV-8. In total, 37,073 samples were tested and 659 (1·78 per cent) were seropositive. The samples came from 5436 herds, of which 45 per cent of herds had only one sample submitted from them. The prevalence was highest in the south of the country, where the outbreak had started, and decreased towards the north. In 340 herds more than 50 per cent of cattle were tested, of which 156 herds were located in infected compartments, and in 37 of these herds (10·9 per cent) at least one positive cow was detected. The average within-herd prevalence in the 37 herds was 39·3 per cent: 2·2 per cent in 11 dairy herds, 68·4 per cent in 20 small-scale herds and 14 per cent in four suckler cow herds. The prevalence differed significantly between herd types but did not show a geographical trend. The average net return for milk production amounted to €2417/cow/year and it decreased significantly on average by €48/cow/year in the bluetongue-infected dairy herds during the bluetongue period. On the small-scale farms, the incidence of mortality increased by 3·2 (95 per cent confidence interval [CI] 1·2 to 9·1) times in the infected herds during the bluetongue period, but the voluntary culling rate decreased by a factor of 2·3 (95 per cent CI 1·1 to 4·8).

IN August 2006, an outbreak of bluetongue virus serotype 8 (BTV-8) was confirmed in the southern part of the Netherlands. The infection was soon confirmed in other areas and in Germany, Belgium, Luxembourg and France (Elbers and others 2007, Mehlhorn and others 2007, Toussaint and others 2007). As part of a European Commission decision, a cross-sectional serological study was carried out in the first half of 2007 to determine the geographical spread of BTV infection in the Netherlands at the end of the first outbreak season. This period was chosen because there would have been little or no activity of the vector *Culicoides* and the level of infection of the ruminant livestock population would have been stable. Cattle were the target species because they were presumed to be the species preferred by biting *Culicoides*.

BTV-8 is thought to induce more limited clinical signs in cattle (Darpel and others 2007, Elbers and others 2007) than in sheep and goats (Backx and others 2007). However, Darpel and others (2007) observed more severe pathology in cattle than was suggested by the mild clinical signs. A study by the Animal Health Service in 2006 of 30 infected and 30 uninfected dairy herds observed fairly limited effects on their health and production (C. J. M. Bartels, personal communication). However, in 2007 farmers reported severe clinical signs in cattle more frequently than had been observed in 2006. This paper reports the seroprevalence of BTV-8 at the animal, herd and regional level in cattle in the Netherlands in the first year of the BTV-8 epidemic, and describes the characteristics and production losses of the infected herds.

MATERIALS AND METHODS

A cross-sectional serological study was carried out between January and June 2007 to determine the geographical spread of BTV infection in cattle in the Netherlands. The epidemiological units for monitoring and surveillance purposes were 21 geographical units called compartments, as proposed in Commission Decision 2005/393/EC (Fig 1). Each compartment was approximately 1000 to 2000 km^2 in area, but because compartment 1 was larger, it was divided into two sub-compartments called 1a and 1b. Blood samples from Dutch cattle submitted to the laboratory of the Dutch Animal Health Service for other voluntary and obligatory health programmes were tested serologically with a competitive ELISA (Institute Pourquier) anonymously and the results reported for each compartment. The ELISA has a sensitivity of approximately 100 per cent and a specificity of more than 99·8 per cent (Toussaint and others 2007). The number of cattle tested in each compartment was dependent, among other factors, on the number of cattle farms, the number of cattle, the number of cattle on bluetongue-infected farms and the presumed expected prevalence of infection in each compartment; the number was calculated to be between 770 and 2040 cattle per compartment.

The within-herd seroprevalence was calculated for herds that had tested at least 50 per cent of their cattle. Analyses were carried out for the infected herds to determine whether the within-herd seroprevalence was significantly different ($P \leq 0{\cdot}05$) for herds of different sizes, different types and different geographical locations. The definitions of the herd types were based on Identification and Registration data of the Dutch cattle sector and the definitions are also used in the Cattle Health Monitor in the Netherlands (Bartels and others 2006, Velthuis and Mourits 2007). Distinctions were drawn between dairy herds, suckler cow herds, traders, beef farms, herds raising youngstock and small-scale herds with fewer than 20 cattle.

In addition, a comparison was made between infected and uninfected herds for several production parameters, such as the net return for 305-day milk production for an average cow in a herd (an indication of a herd's productivity), somatic cell count (SCC), rates of culling and mortality in dairy herds and rates of culling and mortality on small-scale farms, that is farms not producing milk. The data were recorded quarterly from the third quarter of 2002 to the second quarter of 2007. In a multivariate regression model, the bluetongue-infected herds were compared with the uninfected herds with respect to the production parameters before the bluetongue period (the third quarter of 2005 to

Veterinary Record (2008) **163,** 441-444

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TABLE 1: Distribution of the numbers of samples per herd tested for antibodies against bluetongue virus serotype 8 (BTV-8) in the Netherlands in 2006

Number of samples	Number of herds	Percentage of herds
1	2476	45·5
2-10	2297	42·3
11-25	368	6·8
26-50	140	2·6
>50	155	2·9
Total	5436	100·0

the first quarter of 2006) and during the bluetongue period (the third and fourth quarters of 2006). An effect of bluetongue was assumed when there was a significant increase or decrease in the production parameter in the bluetongue herds during the bluetongue period.

The analyses were carried out with a generalised linear model procedure with a correction for repeated measures in STATA 10.0 (XT GEE). The production parameters were the outcome variables and, dependent on the distribution of these parameters (normal, counts of events or binary), the best-fitting distribution (Gaussian, Poisson or binomial) and link function (identity, log or logit) were chosen. The within-herd seroprevalence was modelled as a Poisson distribution (the number of seropositive animals relative to the number tested) with a log-link function. The residuals were investigated to check the model fit and the correctness of the assumptions.

RESULTS

Seroprevalence

In total, 37,073 samples collected from 5436 herds were tested for antibodies to BTV-8, with a range from one to 296 samples per herd (Table 1); 45 per cent of the herds submitted only one sample. Fig 1 shows the 21 compartments and the cattle density in the compartments.

The seroprevalences at the cow and herd level in each of the compartments are shown in Table 2. The within-herd seroprevalence is given for the herds that tested at least 50 per cent of their cattle and could only be determined for the test-positive compartments (compartments 10 to 20). In contrast to the proportion of infected herds that showed a clear trend (increasing from north to south) the within-herd seroprevalence did not show this trend (Fig 2).

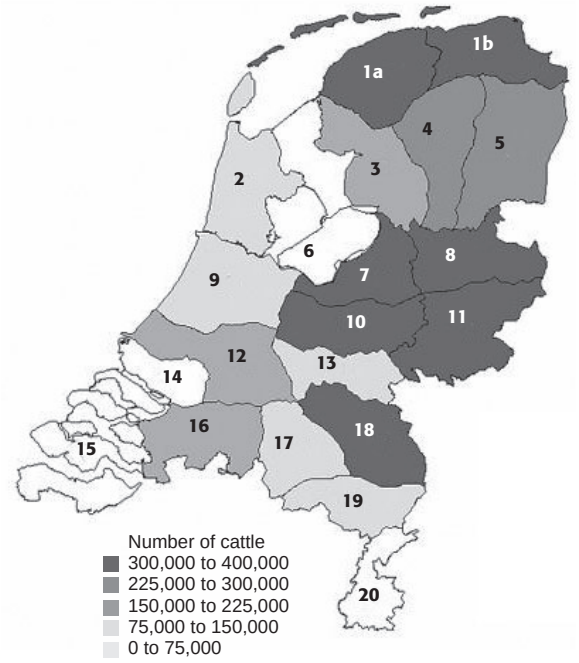

FIG 1: Twenty-one compartments designated for the cross-sectional study of the seroprevalence of bluetongue virus serotype 8 in the Netherlands and the numbers of cattle per compartment

Table 3 shows the distribution of samples across the different herd types; 48 per cent of the samples came from dairy herds, 26 per cent came from suckler herds and 15 per cent came from small-scale farms; 127 herds could not be typed in one of the six type categories. The numbers of samples from dairy herds and traders were as expected, the numbers from suckler herds and herds raising youngstock were higher than expected and the numbers from the other types of herds were lower than expected (Table 3).

In 340 of the herds, more than 50 per cent of the cattle were tested; 62 of them were dairy herds and 205 were

TABLE 2: Seroprevalence of bluetongue virus serotype 8 (BTV-8) at the cattle and herd level in 21 compartments in the Netherlands

Compartment	Number of seropositive animals	Total number of animals tested	Seroprevalence (%)	Number of herds that tested >50 per cent of their cattle*	Number of seropositive herds	Within-herd seroprevalence (%)
1a	0	2221	0	–	–	–
1b	0	1892	0	–	–	–
2	4	1989	0·2	–	–	–
3	0	2073	0	–	–	–
4	0	2121	0	–	–	–
5	1	2090	0	–	–	–
6	0	1197	0	–	–	–
7	2	2012	0·1	–	–	–
8	1	2043	0	–	–	–
9	3	2099	0·1	–	–	–
10	8	2031	0·4	28	2	1·4
11	2	2093	0·1	22	1	2·3
12	10	2096	0·5	18	0	–
13	18	2021	0·9	19	4	0·3
14	14	547	2·6	3	1	8·3
15	75	789	9·5	7	3	31·3
16	52	2047	2·5	9	3	1·0
17	84	2053	4·1	24	12	27·4
18	33	2042	1·6	16	5	2·9
19	36	776	4·6	2	1	0·2
20	316	841	37·6	8	5	50·0
Total	659	37,073	1·8	156	37	39·3

* Only for test-positive compartments in which at least two test-positive herds tested more than 50 per cent of their cattle

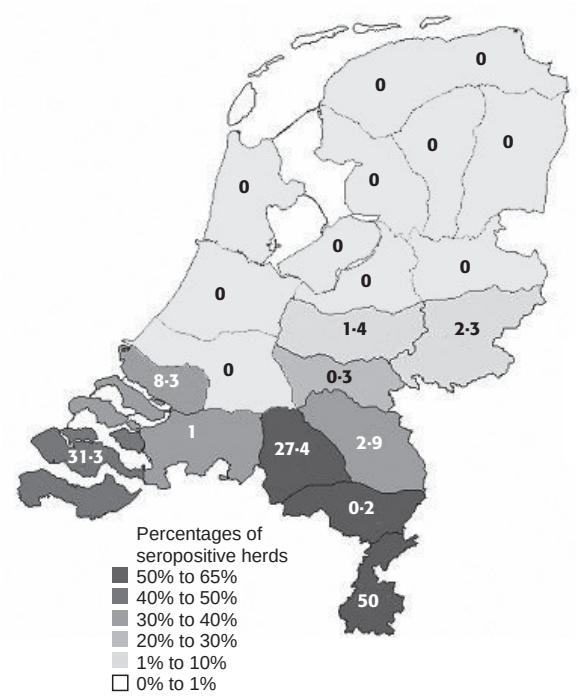

FIG 2: Percentages of herds that tested seropositive for bluetongue virus serotype 8 and the within-herd prevalence (shown in each compartment) in 21 compartments in the Netherlands

small cattle holdings. In 37 of them (10·9 per cent) at least one positive cow was detected; 11 of these herds were dairy herds, 20 were small-scale farms, four were suckler herds, and there was one veal calf farm and one beef farm (Table 4). Fig 3 shows the percentage distribution of the within-herd seroprevalence among these 37 infected herds; 40 per cent of them had a within-herd seroprevalence below 10 per cent, and approximately 20 per cent had a seroprevalence above 90 per cent.

Table 4 gives the within-herd seroprevalence for the different herd types. The average seroprevalence in the 37 herds was 39·3 per cent: 2·2 per cent in the 11 seropositive dairy herds, 68·4 per cent in the 20 small-scale herds and 14 per cent in the four suckler herds.

TABLE 3: Distribution of different types of cattle holdings in the Netherlands in 2006 and the distribution in the cross-sectional sample

Herd type	Distribution in 2006 Number (%)	Distribution of cross-sectional sample Number (%)
Traders	290 (0·64)	41 (0·75)
Farms raising youngstock	1153 (2·54)	235 (4·32)
Small-scale farms	14,286 (31·4)	819 (15·1)
Dairy herds	20,897 (46·0)	2602 (47·9)
Herds of uncertain type	338 (0·74)	127 (2·34)
Beef herds	3355 (7·38)	197 (3·62)
Suckler cow herds	5153 (11·3)	1415 (26·0)
Total	45,472 (100·0)	5436 (100·0)

TABLE 4: Numbers and percentages of different types of cattle herds that were positive for bluetongue virus serotype 8 (BTV-8) and the mean (sd) within-herd prevalence in the infected herds that sampled at least 50 per cent of their cattle

Herd type	Number of seronegative herds	Number of seropositive herds	Percentage of seropositive herds	Within-herd seroprevalence
Traders	13	0	0	–
Farms raising youngstock	2	0	0	–
Small-scale farms	185	20	11	68·4 (31·5)
Dairy herds	51	11	22	2·2 (1·9)
Herds of uncertain type	7	0	0	–
Beef herds	9	2	22	2·9 (2·8)
Suckler cow herds	36	4	11	14·0 (12·0)

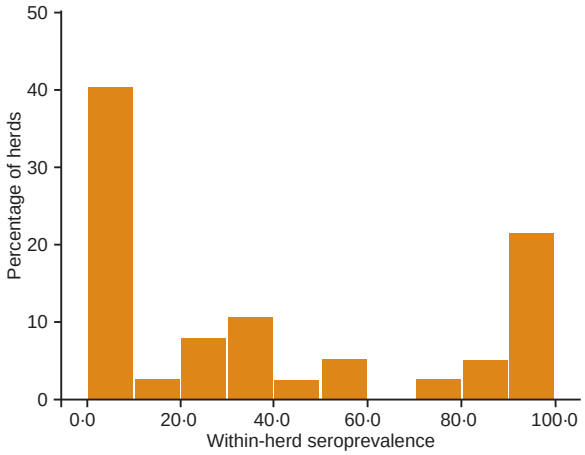

FIG 3: Distribution of within-herd seroprevalence of bluetongue virus serotype 8 in the 37 infected Dutch herds in which at least 50 per cent of cattle were sampled

The results of the statistical analysis of the within-herd seroprevalence are shown in Table 5. It was significantly higher in the small-scale farms than in the dairy farms, significantly higher in the southern part of the Netherlands than in the northern and central parts, and increased with herd size. The square and square root of herd size was tested but was not significant, and the plot of the residuals with herd size showed a linear association.

Production losses

The average net return for 305-day milk production was €2417 per cow. It decreased significantly by €48 per cow (95 per cent confidence interval [CI] €3 to €94) in the bluetongue-infected dairy herds during the bluetongue period compared with the period before the outbreak. The other production parameters were not affected significantly.

On the small-scale farms, the incidence of mortality increased by a factor of 3·2 (95 per cent CI 1·2 to 9·1) in the bluetongue-infected herds during the bluetongue period, but the voluntary culling rate decreased by a factor of 2·3 (95 per cent CI 1·1 to 4·8) compared with the period before the outbreak.

DISCUSSION

The blood samples came from herds that participated in voluntary and/or obligatory health programmes, and this selection may have adversely affected the representativeness of the sample. However, they had the advantage that they were not submitted because of a suspicion of BTV-8, which might have resulted in an overestimation of its prevalence. Although it was not a random sample, the cross-sectional sample was fairly representative of the distribution of types of cattle herd in the Netherlands. Small-scale herds were slightly under-represented and suckler herds were over-represented. However, the authors consider that the estimates of prevalence are accurate for the Netherlands at the end of the first outbreak season. The prevalence was low at both the compartment level and the herd level. There were only a few infected herds, but the results indicated that the dairy herds had a lower within-herd seroprevalence than the small-scale herds and suckler herds, and that smaller herds had a lower within-herd seroprevalence than larger herds. Furthermore, the within-herd seroprevalence was significantly higher in the southern part of the Netherlands than in the central and northern parts; this was to be expected because the outbreak started in the south and the infection pressure was therefore

TABLE 5: Results of the generalised linear model for the within-herd seroprevalence of bluetongue virus serotype 8 (BTV-8) in 306 herds in the Netherlands

Variable	Coefficient (β)	SE	P	Exp (β)	Lower limit	Upper limit
Intercept	–6·54	0·90	0·00	–	–	–
Small-scale farms	1·89	0·76	0·01	6·60	1·50	29·07
Suckler cow herds	0·44	0·67	0·51	1·55	0·42	5·68
Dairy farms	0·00	–	–	1	–	–
Herd size (cows)	–0·008	0·003	0·05	0·99	0·99	1·00
South Netherlands	3·57	0·45	0·00	35·55	14·83	85·22
North and central Netherlands	0·00	–	–	1	–	–
DScale (based on the deviance)	0·68					

higher there. The higher seroprevalences in the small-scale herds and suckler herds may be a result of differences in housing and grazing management relative to the dairy herds. Most dairy herds graze their cattle for a limited period during the day (between milkings) and keep their cattle in overnight, whereas small-scale herds and suckler herds usually graze their cattle outside for 24 hours a day. Furthermore, the cattle in the small-scale herds and suckler herds are more likely to have grazed more extensively in areas that might be more favourable for the vector, and so have been more exposed to it.

In 2006, bluetongue appeared to have had an effect on milk production because in the dairy herds the net returns decreased by 2 per cent. The validity of this result may be limited because there were only 11 infected dairy herds in the study and the average seroprevalence among them was only 2.2 per cent. In the 20 infected small-scale herds, mortality was increased during the bluetongue period compared with the same period a year earlier. However, these herds each kept fewer than 20 animals, and as a result only a small number of cows would have died due to bluetongue.

The results of this study depict the situation in the Netherlands in 2006, when BTV-8 was an emerging infection. Similar results may be observed at the start of a BTV-8 epidemic in other naive populations.

ACKNOWLEDGEMENTS

The study was partly funded by the European Union, the Dutch Ministry of Agriculture, Nature and Food Quality and the Dutch Livestock Industry. The authors would like to thank Bert de Roo for the provision of test data used in the study.

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